Confining safflower pollen during regeneration of germplasm seed stocks

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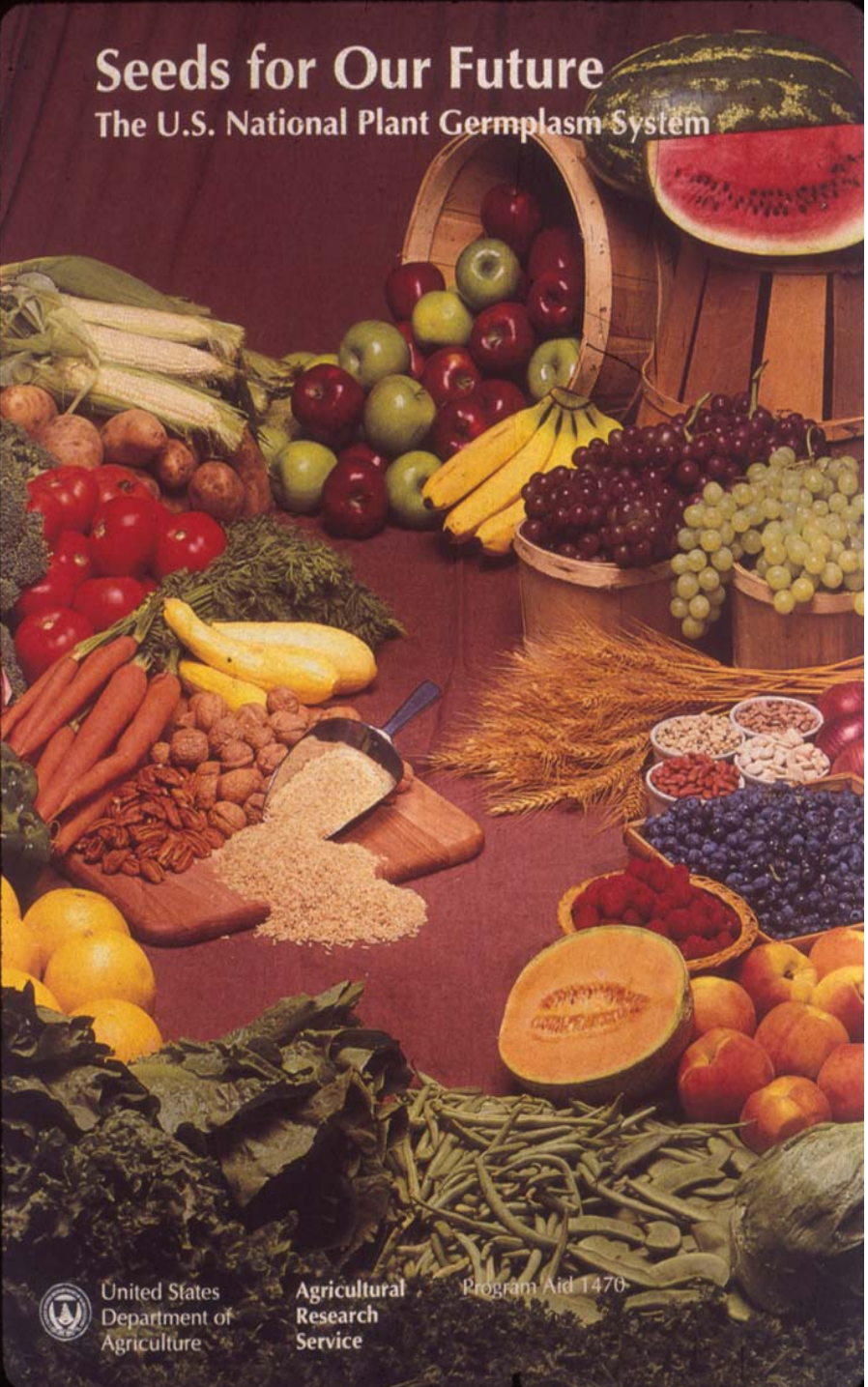
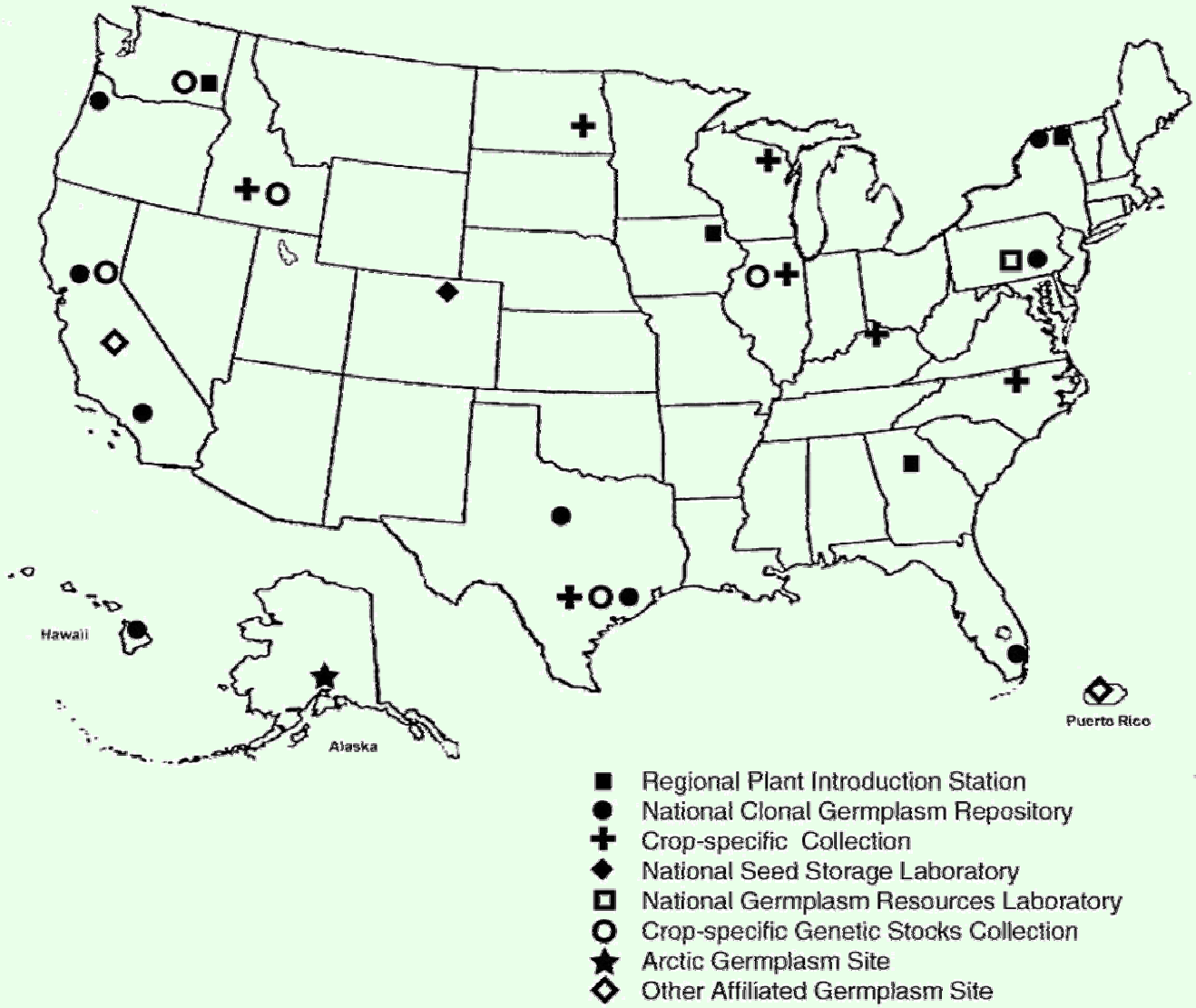
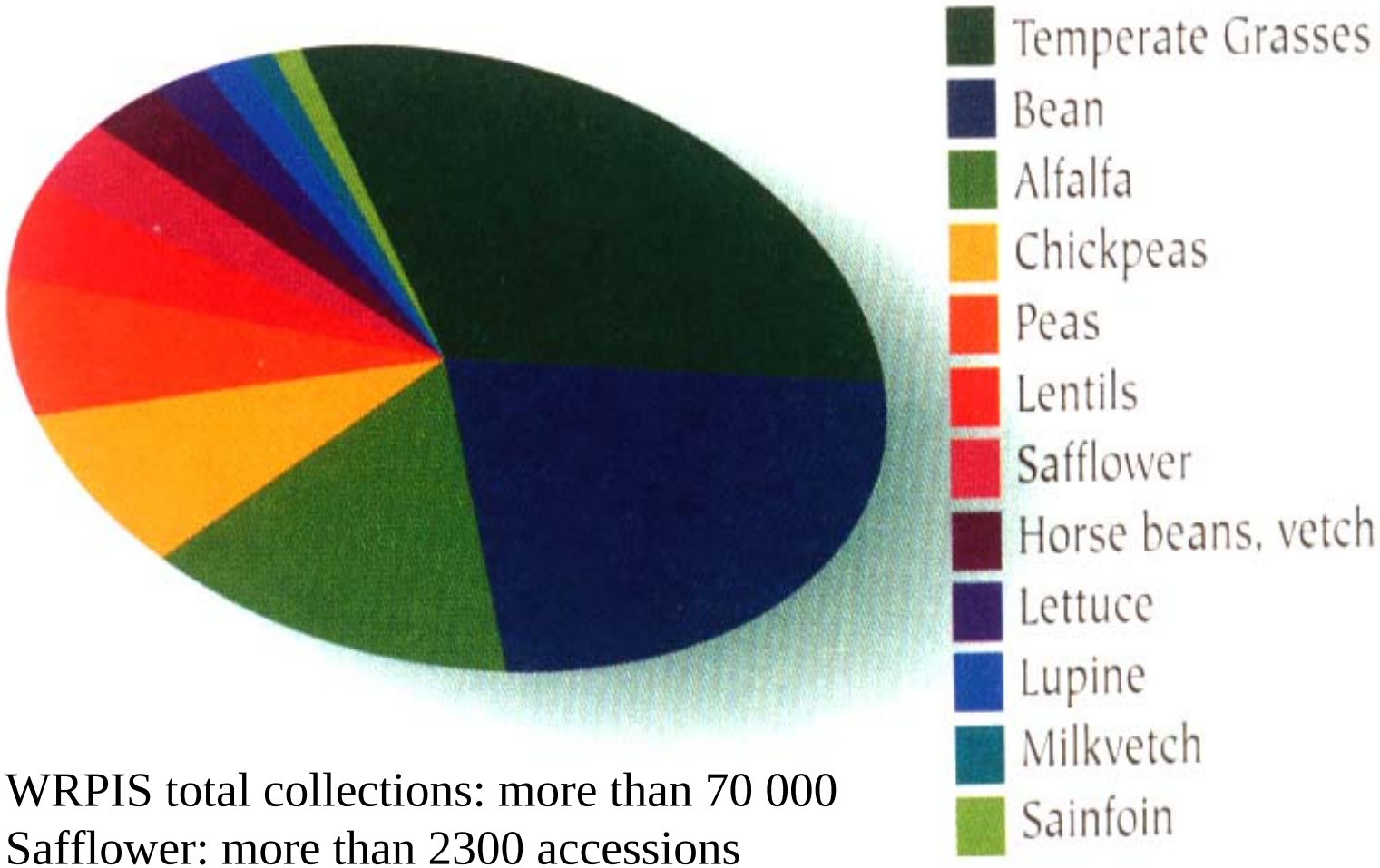


- **About 20 sites with "active collections," focused on seed or clonally-propagated crops.**
- **Active sites acquire, maintain, regenerate, distribute, document, characterize, evaluate.**
- **Associated research.**
- **"Base collection"; preservation research.**
- **GRIN database: www.ars-grin.gov**
- **Crop Germplasm Committees; University, NGO, industry cooperators, and ARS.**



The U.S. National Plant Germplasm System

WRPIS Major Collections



WRPIS total collections: more than 70 000
Safflower: more than 2300 accessions


Distribution: about 20,000 total samples per year, one-third outside US.

Safflower accession 543980 from China

Outline

- Safflower pollination biology
- Reported outcrossing rates
- Outcrossing agents
- Genetic marker systems
- Outcrossing to wild relatives
- Pollen confinement in regeneration of genetic stocks

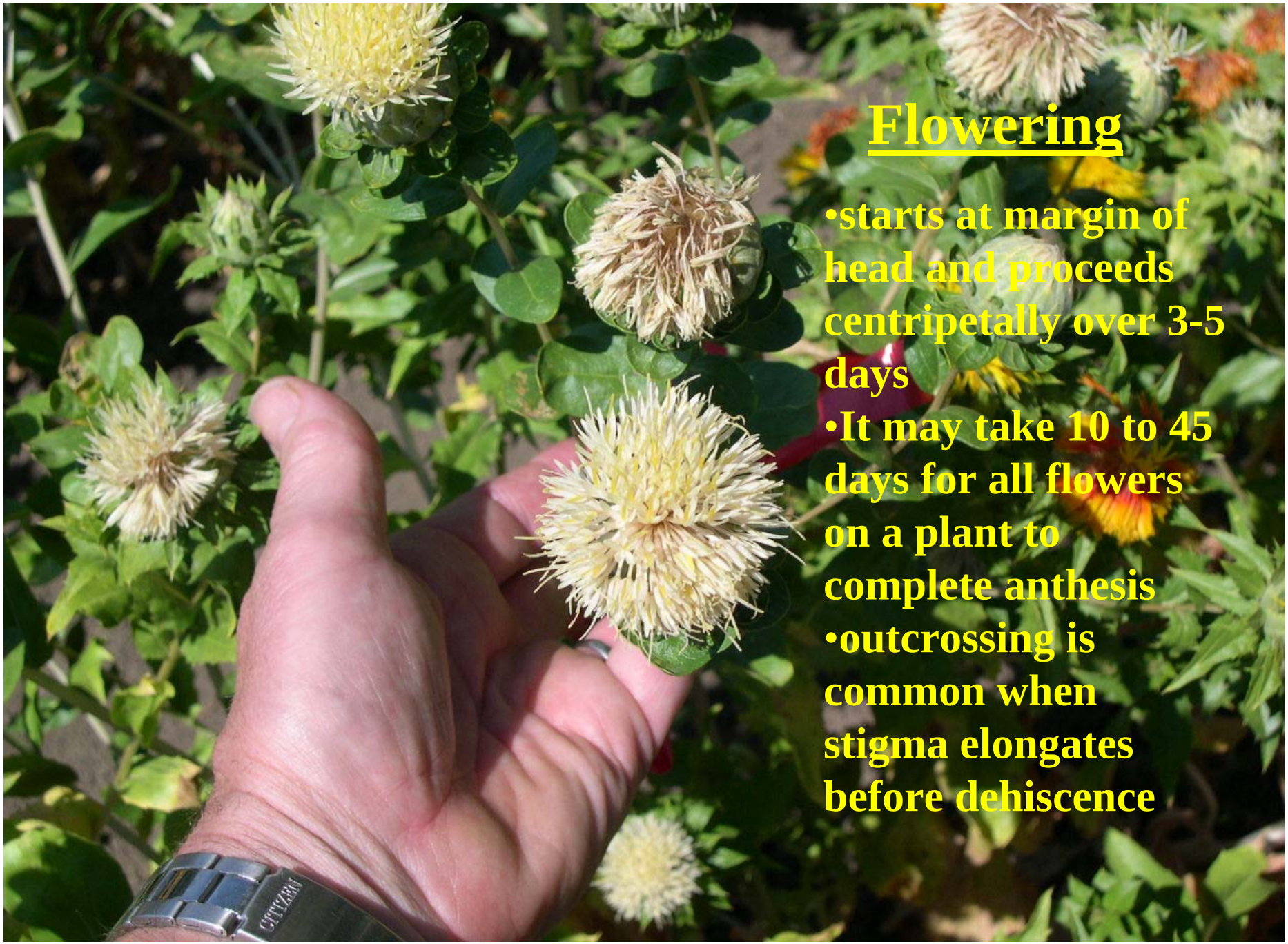

Safflower Pollination Biology

Heads

- 15 to 150 per plant
- 1.2 to 4 cm diameter
- 20 to 100 florets per head

Flowers

- tube like with a five pointed tip
- anthers united into tube
- filaments free and attached to corolla
- style surrounded by anther tube



Flowering

- starts at margin of head and proceeds centripetally over 3-5 days
- It may take 10 to 45 days for all flowers on a plant to complete anthesis
- outcrossing is common when stigma elongates before dehiscence

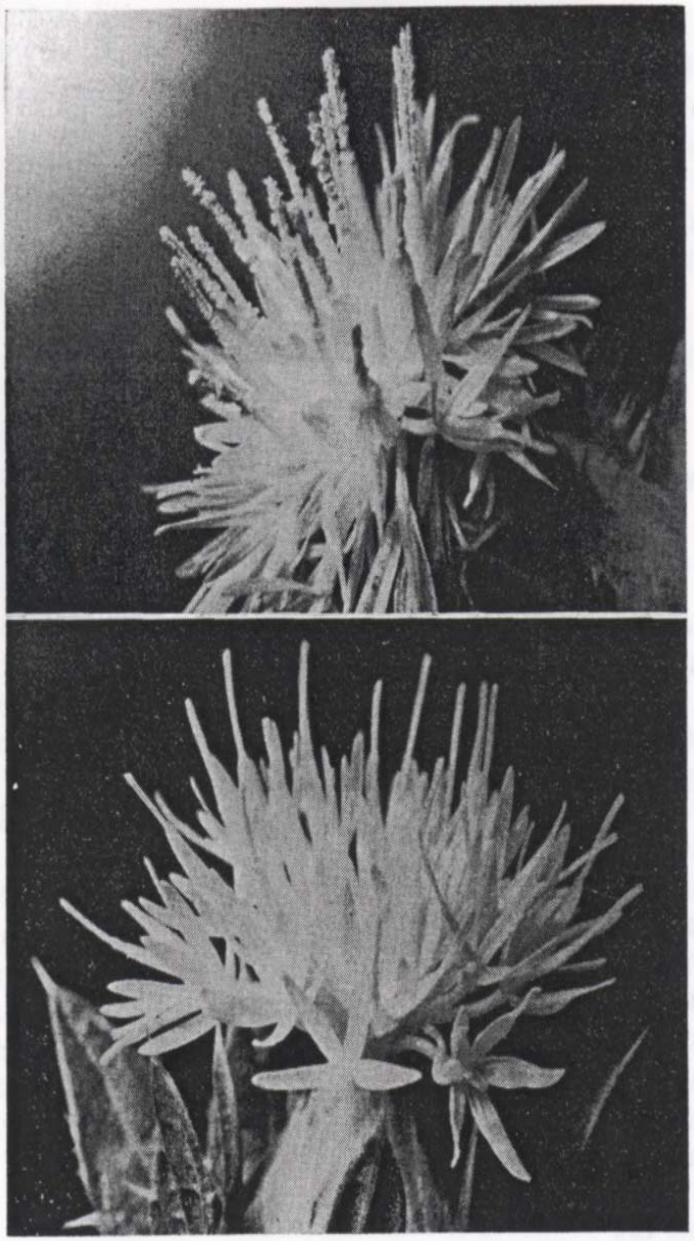


Figure 1. Stigmas covered with pollen in Line A (top) but free of visible pollen in Line B (bottom).

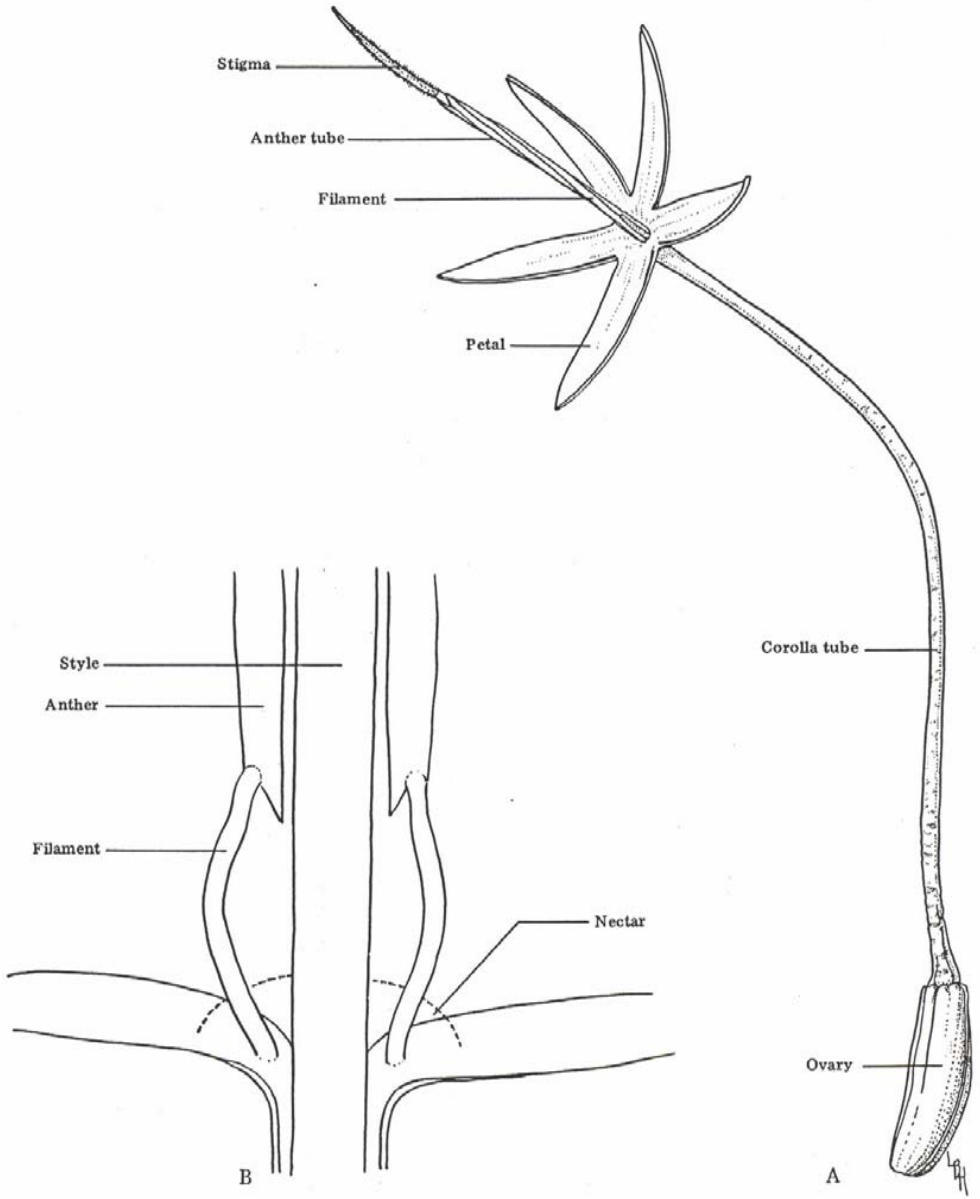


FIGURE 173.—Longitudinal section of safflower floret. *A*, Floret, ×6; *B*, filaments and adjoining area, greatly enlarged.

Natural crossing in five open pollinated safflower lines grown between rows of a dominate tester*

		Natural Crossing %	
Line No.	No. Plants	Average	Range
33	9	13.3	9.8-19.4
34	55	19.8	4.9-83.0
48	20	28.8	9.0-93.6
49	19	15.1	6.8-54.5
50	19	17.1	0.0-59.6

*data from Claassen, 1950

Outcrossing rates

- Can vary widely; from zero to near 100%
- Dependent on genotype and insect activity
- Average is usually between 15 and 20% based on dominant flower color markers (Claassen, 1950 and others)
- No allowance for sibling crossing or insect color preference
- Plants with little outcrossing may be fertilized early in the day before insects are active
- Kadam & Pantaka (1942) reduced outcrossing to “negligible” with separation of 30 m interspaced with wheat or other crops

Genetic markers: more work needed?

- Flower color in order of dominance: yellow, orange, red, yellow, and white. Most if not all work to date. Possible interactions with bee color preference.
- Spiny (dominant) and non-spiny (recessive)
- Seed hull dominant white (StpStp) and recessive gray strip (stpstp).
- High linoleic, low oleic (OLOL) and low linoloeic/high olieic (olol)
- Co-dominant molecular markers; isozyme variation reported by Carapetian (1994).

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5/
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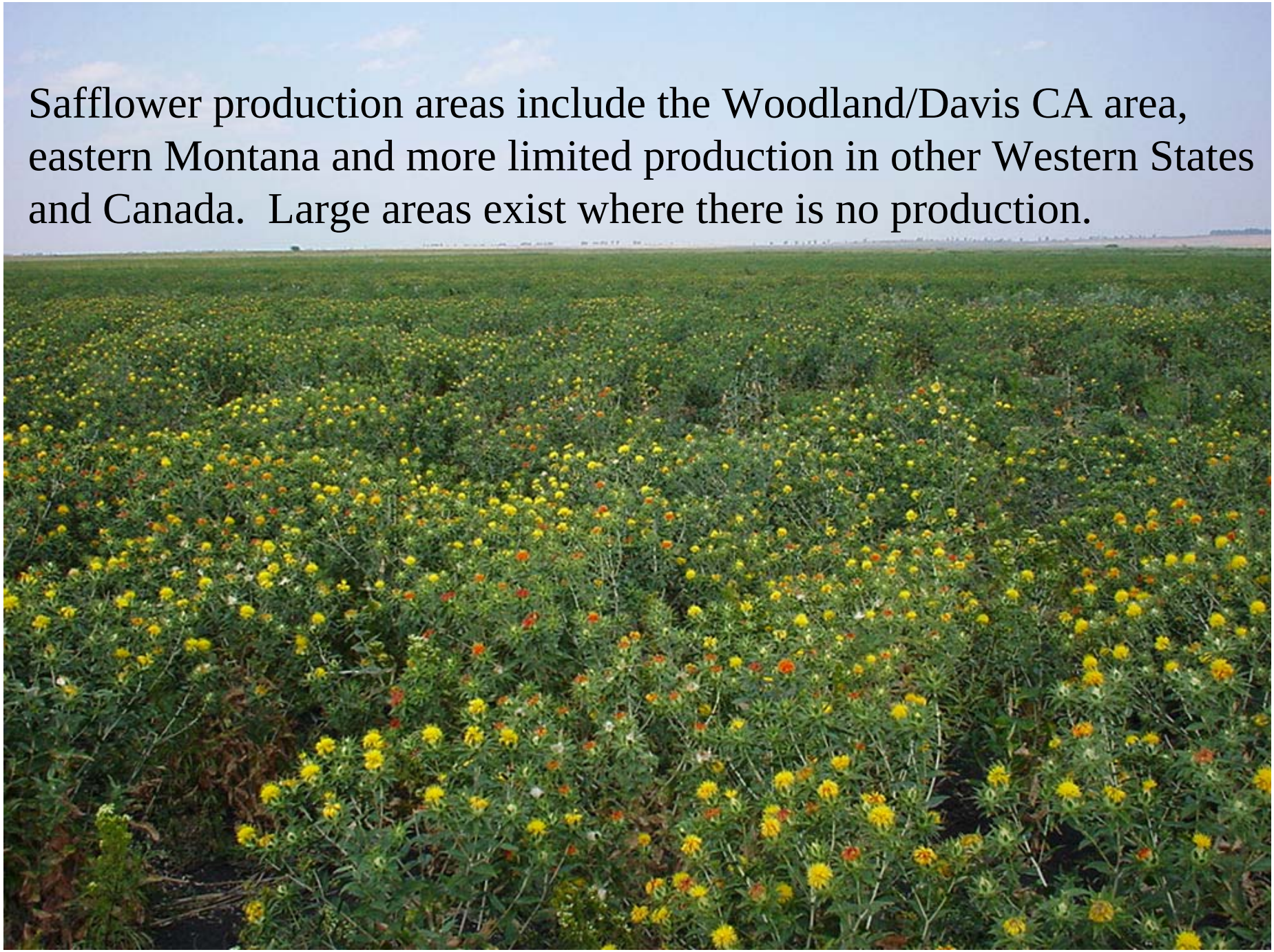
VALUE

Outcrossing agents

- Forty species of native bees were collected on safflower blossoms in Arizona, but populations were small compared to honey bees (Butler et al., 1966).
- Wind does not appear to be a significant factor in outcrossing (Claassen, 1950)
- Although selfing is predominate, reduced seed production is possible in the absence of pollinators (Boch, 1961)

Safflower production areas include the Woodland/Davis CA area, eastern Montana and more limited production in other Western States and Canada. Large areas exist where there is no production.

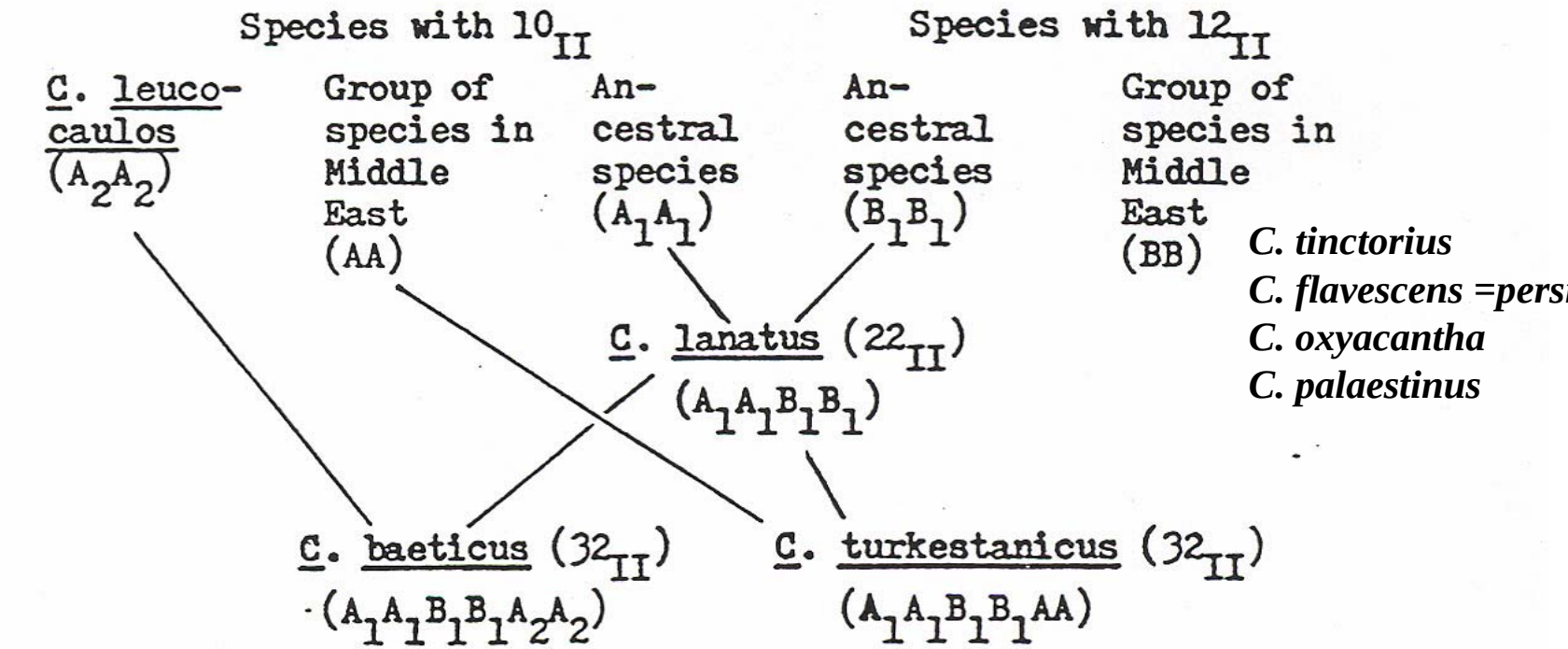


Safflower Outcrossing to Other Species

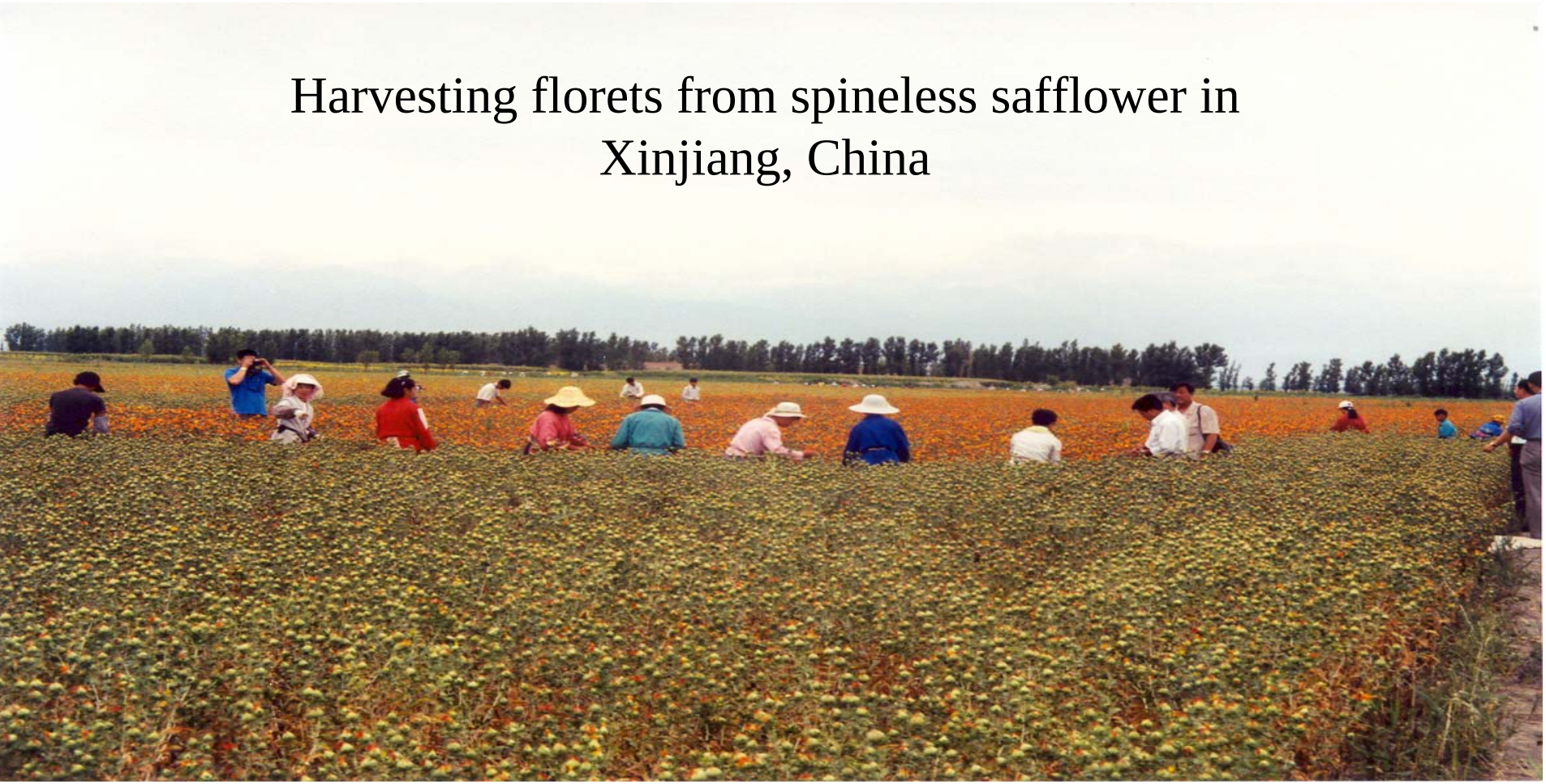
- About 25 species of wild safflower divided into four sections based on chromosome number (Ashri & Knowles, 1960). Many are weedy.
- Safflower (*C. tinctorius*) has n=12 chromosomes and will cross readily with other species with n=12 including the noxious weed *C. oxyacnathus*.
- The distribution of weedy *Carthamus* is very limited in Western North America
- McPherson et al. (2004) concluded that extensive New World locations could be found were weedy *Carthamus* would not be present.

After Ashri & Knowles, 1960

A summary of assumed species relationships is as follows (proposed genomic formulas are in parentheses):



Harvesting florets from spineless safflower in Xinjiang, China



Regeneration of genetic stocks at the WRPIS

- Needed to provide high quality seed for researchers
- Usually grow about 300 accessions per year for seed increase
- Based on caging accessions to exclude insects and prevent outcrossing to other accessions

Problems

- Initially (1960's-1970's) cotton bags were used with only 3-5 plants per accession covered. Seed distribution of outcrossed seed.
- This was detrimental to seed quality and quantity and low populations reduced genetic diversity within accessions
- In 1980's screen bags were used to cover entire populations, helping to maintain diversity, but seed quality was still questionable
- Both cotton and screen bags plants tended to result in plants being packed together, suggesting high humidity was a factor in the poor seed quantity and quality.

Solution

- Screen cages were constructed to enclose each accession with minimal packing
- Seed production is adequate and germination generally above 90%
- Some accessions with high levels of natural outcrossing may require bees in screen cages, such a Rubis's thin-hull (male-sterile)
- Some wild relative such as *C. oxyacanthus* and *C. flavescens(=persicus)*, with self-incompatibility mechanisms, would also require pollinators

Summary

- Reports of safflower outcrossing rates average 15 to 20%, but can be much higher depending on insect activity and genotype
- Crosses to wild *Carthamus*,especially those with n=12 chromosomes, could occur if plants are in close proximity
- However, there are large areas of North America with neither safflower production nor wild *Carthamus*
- Isolation of safflower in suitable growing environments should be possible.


Questions?